# GC-MS Study on the Aroma of Thirteen Egyptian Mango Cultivars

Mokhtar M. Bishr<sup>1</sup>, Mohamed A. El-Degwy<sup>2</sup> and Sayed A. Mossa<sup>2</sup>

Abstract: The Egyptian cultivars of Mangiferaindica L., have a great distinct difference in their aroma and fragrance, and due to the importance of the aroma (essential oils) in pharmaceutical, food, cosmetics and perfumes industries, it was necessary to investigate the composition of the essential oil of these cultivars. Gas chromatography Mass spectrometry using head space was carried out for the investigation of the composition of the essential oil present in the peel of thirteen Egyptian cultivars. The identification depends on the retention time, mass spectrum and data obtained from Wiley library. In order to find out the reason for the differences between these cultivars, the major common components, as well as the unique components present in each oil were investigated. In the near future, a detailed study for each separate cultivar will be done.

Keywords: Mango cultivars, GC/MS,

### I. Introduction

Mango (Mangiferaindica, L.) is a perennial tree belonging to the family Anacardiaceae. This plant is native to Southeastern Asia and afterward spread to other parts of tropical world (1-6).

Anacardiaceae is a family comprising 70 genera; one of these genera is Mangifera. Most Mangifera(Mango) in Egypt was said to be of Indian origin(4&7). However, enough time has passed since the first crop was introduced to permit a tradition of cultural methods to develop in Egypt and to permit the development of a large number of common seedlings of diverse types (4&8).

This fact is clear now as most of the Egyptian cultivars are polyembryonic, while most of the Indian cultivars are monoembryonic (4&8).

It is also expected that the Egyptian cultivars have come from Southern coastal region of India, where polyembryonic types are found (4&9). Another expected source is the coastal region of East Africa and Kenya, where many polyembryonic cultivars of Mango are grown (4&10).

The different cultivars of Egyptian Mango have great and distinct differences in their appealing aroma and fragrance, where we can find wide range of sweet strong spicy flavors. Therefore, we decided to carry out the present comparative GC/MS study on the essential oil (aroma) content in the peel of 13 cultivars grown in Egypt.

Although, a large number of components were detected in all oils; but we chose only the common major components as well as the unique components in each oil for the present study.

Finally, to the best of our knowledge, this is the first study on the chemical composition of essential oils of 13 cultivars using GC/MS with head space.

Hydro-distillation method was avoided as it is more tedious, time consuming, needs several kilograms of the peel and hydrolysis by hot water as well as loss of volatile components is expected; whereas in the present study, small piece of the peel was required.

#### 2.1. Plant material:

### II. Materials And Methods

Fruits of thirteen mango cultivars were purchased from the Mango farms in Ismailia and Sharkyah, Egypt and identified by field experts in each farm, where their opinions were found to be identical. Third sample for each cultivar was purchased from the local market. A piece of the peel of the three samples of each cultivar was used separately for the GC/MS analysis.

These cultivars, arranged alphabetically, are: Alfons, Baladi, Ewaise, Fagr, Fuss, Ket, Langary, Misk, Naomee, Sobaa El-set, Succari, Tomy and Zebda.

### 2.2 Equipments

Gas Chromatography- Mass Spectrometry:Shimadzu GC-MS, Model QP-2010 Ultra, equipped with head space AOC-5000 auto injector is used for the analysis of essential oil present in the peel of the above mentioned Mango cultivars.

# 2.3 Experimental:

- **2.3.1** Sample preparation: A small piece of one square centimeter, from the fresh peel of three different samples; (two from two different farms: Ismailia &Enshas,Sharkyah and the third from the local market), for each cultivarwas used directly for the GC/MS analysis.
- **2.3.2** GC/MS analysis: The GC/MS analysis was carried out using Shimadzu GC-MS, Model QP-2010 Ultra, equipped with head spaceAOC-5000 auto injector, under the following condition:
- Column: Rtx-5 MS 30 meter length, 0.25mm ID, 0.25 um film thickness(Cross bond5% diphenyl/95% dimethyl polysiloxane).
- Detector: QP2010 ultra Mass spectrometer.
- Carrier gas: Helium.
- Oven temperature program:
- Start temperature =  $40^{\circ}$ C hold time = 2 minutes
- Final temperature  $= 210^{\circ}$ C hold time = 5 minutes
- Rate  $= 5^{\circ}C/$  minute
- GC-MS program:
- Ion Source temperature  $= 230^{\circ}C$
- Interface temperature  $= 230^{\circ}C$
- Solvent cut Time = 2 minutes.
- Detector gain mode = relative.
- Detector Gain = 1.08 kv + 0.00 kv.
- Run time = 41 minutes.
- ACQ mode = Scan
- Event time = 0.3 sec.
- Scan speed = 1.666
- Start m/z = 35.00
- End m/z = 500.0

### III. Results And Discussion

The results gained from the three samples of each cultivar were one and the same in the GC-MS analysis. The analyzed cultivars gave different number of the components present in their essential oil as follow: Alfons (76 components), Baladi (74 components), Ewaise(19 components), Fagr (55 components), Fuss (32 components), Ket (67 components), Langary (50 components), Misk (71 components), Naomee (33 components), Sobaa El-set (17 components), Succari(47 components), Tomy (45 components) and Zebda (23 components). Details of the presence of various essential oil components with their percentage in different tested cultivars are shown in the following table (1).

Table (1): GC-MS analysis of essential oil content in the peel of 13 Mango cultivary
--

No.	Component	Rt,(min)	No. of Cultivars	Percentage of its presence in different cultivars
1	Beta Myrecene	10.065	12	Baladi(50.83), Tomy(2.27), Fuss(2.14), Ewaise(1.23) Alfons(1.22), Langary(1.21), Naomee(1.20), Zebda(1.12), Sobaa el-set(1.11), Fagr(1.10), Succari(1.00), Misk(0.71).
2	2-Furan-carboxaldehyde	5.366	12	Succari(32.06), Ewaise(30.29), Ket(26.34), Fagr(25.08), Naomee(12.56), Zebda(12.30), Langary(11.14), Misk(3.25), Tomy(2.60), Sobaa el-set(2.49), Baladi(2.07), Alfons(1.46).
3	Ethanone,2,2-dimethoxy-1,2 diphenyl	34.067	12	Ket(4.46), Ewaise(3.87), Naomee(3.20), Zebda(2.60), Misk(1.46), Langary(1.28), Baladi(1.23), Succar(1.10), Fagr(1.00), Sobaa el-set(0.98), Alfons(0.55), Tomy(0.47),.
4	Delta-3-carene	10.67	10	Tomy(57.19), Naomee(35.39), Langary(34.05), Ket(32.66), Fuss(7.19), Fagr(6.41), Zebda(6.01), Ewaise(4.40), Baladi(0.34), Misk(0.26).
5	Nonanal	13.672	10	Baladi(2.63), Fuss(2.62), Ewaise(2.15), Naomee(1.05), Zebda(0.96), Ket(0.86), Fagr(0.85), Langary(0.38), Misk(0.19), Alfons(0.10).
6	Trans-caryophyllene	23.011	10	Misk(4.81), Ket(2.72), Fuss(2.22), Langary(1.50), Naomee(1.11), Baladi(1.01), Zebda(0.85), Fagr(0.53), Tomy(0.200), Alfons(0.19).
7	2-furancarboxaldehde, 5- methyl.	9.219	10	Succari(6.13), Ket(2.36), Fagr(1.96), Langary(0.900), Misk(0.800), Naomee(0.62), Sobaa el-set(0.24), Tomy(0.22), Baladi(0.21), Alfons(0.100).

# GC-MS Study on the Aroma of Thirteen Egyptian Mango Cultivars

				Tomy(19.02), Ewaise(1.20), Zebda(0.91), Ket(0.63),
8	Alpha-pinene	8.234	10	Langary(0.64), Misk(0.500), Sobaa el-set(0.44), Naomee(0.42), Baladi(0.41), Fagr(0.36).
9	Alpha-terpinolene	13.168	9	Zebda(49.18), Ewaise(30.25), Fuss(26.85), Fagr(16.68), Naomee(10.55), Tomy(2.75), Langary(1.83), Ket(1.59), Baladi(1.58).
10	dl-lemonene	11.247	9	Tomy(3.10), Fuss(2.70), Zebda(2.10), Naomee(1.62), Fagr(1.28),Langary(1.22), Ket(1.02), Baladi(0.21), Misk(0.21).
11	Alpha-copaene	21.789	9	Naomee(4.57), Misk(3.84), Alfons(3.56), Baladi2.92), Fuss(1.91), Ket1.84), Fagr(.47), Langary(1.340, Tomy(0.28).
12	2-heptadecanone	34.221	9	Langary(13.75), Fagr(3.00), Zebda(1.98), Naomee(1.97),Baladi(1.77), Ket(1.17), Tomy(1.14), Soba el-set(1.13), Misk(0.73)
13	Linalool oxide	12.689	9	Ewaise(4.02), Fagr(3.87), Zebda(1.62), Ket(0.83), Langary(0.80), Naomee(0.450), Misk(0.38), Tomy(0.29), Baladi(0.23).
14	Hexadecanal	30.137	9	Ket(1.82), Alfons(1.27), Baladi(1.07), Naomee(1.04), Misk(0.71), Fagr(0.500), Langary(0.34), Succari(0.32), Tomy(0.100).
15	Cis-methyl propenylketon	3.389	9	Zebda(1.90), Langary(0.600), Ket(0.59), Baladi(0.42), Misk(0.37), Succari(0.29), Soba el-set(0.21), Tomy(0.19), Alfons(0.15).
16	Octanoic acid ethyl ester	16.559	8	Zebda(3.46), Naomee(2.26), Baladi(2.35), Alfons(2.19), Langary(1.75), Fuss(1.49), Succari(1.22), Misk(0.23).
17	l-phellandrene	10.473	8	Zebda(1.17), Tomy(0.87), Ket(0.63), Naomee(0.62), Langary(0.44), Baladi(0.41), Fagr(0.39), misk(0.15).
18	Germacrene-D	24.649	8	Ewaise (2.29), Alfons(2.22), Baladi(2.02), Fuss(1.47), Langary(0.75), Misk(0.71), Zebda(0.700), Fagr(0.24)
19	Butanoic acid ethyl ester	4.591	8	Alfons(15.32), Baladi(7.23), Fuss(3.95), Langary(3.42),
20	Benzene acetaldehyde	11.786	8	Naomee(0.57), Ket(0.45), Tomy(0.27), Misk(0.15). Ewaise(1.41), Naomee(1.04), Langary(0.38), Ket(0.33), Misk(0.27), Fagr(0.25), Baladi(0.14), Alfons(0.13).
21	Alpha-terpineol	16.41	8	Fagr(2.59), Fuss(2.23), Tomy(1.05), Langary(0.67), Naomee(0.46), Misk(0.25), Ket(0.23), Baladi(0.200).
22	N-heptanal	7.299	7	Zebda(0.82), Ket(0.77), Naomee(0.64), Baladi(0.46), Fagr(0.39), Misk(0.26).Soba el-set(0.22)
23	Ethanone,1-(2-furanyl)	7.622	7	Succar(1.80), Ket(1.38), Naome(1.02), Fagr(0.66), Misk(0.59), Langary(0.53), Tomy(0.09).
24	Linalool	13.544	7	Fagr(5.23), Naomee(1.38), Langary(0.99), Baladi(0.96), Misk(0.39), Ket(0.29), Tomy(0.11).
25	Alpha-humulene	23.932	7	Misk(2.23),Fuss(1.34), Ket(1.13), Langary(0.59), Baladi(0.46), Fagr(0.26), Alfons(0.12).
26	Alpha-terpinene	10.853	7	Fuss(2.90), Ket(1.09), Tomy(0.65), Fagr(0.62), Naomee(0.51), Langary (0.29), Baladi(0.11).
27	1,3,6-octatriene,3,7- dimethyl,€.	11.886	6	Sobaa el-set(23.32), Misk(12.76), Fagr(1.16), Baladi(0.900), Alfons(0.1), Tomy(0.1).
28	Camphene	8.683	6	Ket(0.700), Succari(0.67), Tomy(0.59), Misk(0.49), Baladi(0.19), Soba el-set(0.17).
29	1-heptadecanol	32.339	6	Alfons(2.53), Ewaise(1.81), Succari(1.08), Langary(0.42), Fagr(0.36), Baladi(0.22).
30	Beta-citronellol	16.805	6	Fuss(2.72), Fagr(0.46), Ket(0.36), Succari(0.300), Baladi(0.25), Misk(0.23).
31	Geraneol	18.256	5	Fuss(2.67), Fagr(0.600), Ket(0.31), Baladi(0.13), Tomy(0.10).
32	Alpha-cedrol	21.813	5	Succari(26.50), Ewaise(5.72), Zebda(4.75), Soba el- set(0.65), Alfons(0.45).
33	2-butenoic acid ethyl ester	5.672	5	Fuss(5.55), Baladi(3.14), Langary(2.26), Alfons(0.93), Tomy(0.12)
34	2-propenoic acid, 2- methylethyl ester	4.926	5	Langary(3.70), Naomee(2.65), Baladi(0.85), Tomy(0.21), Alfons(0.19).
35	Cis-ocimene	11.576	5	Soba el-set(65.9), Misk(38.25), Fagr(0.67), Alfons(0.51), Baladi(0.13).
36	2-propanone-1-hydroxy	3.053	5	Langary(2.15), Ket(1.34), Tomy(0.63), Misk(0.42), Alfons(0.12).
37	Borneol	15.368	5	Fuss(2.25), Succari(1.74), Misk(0.29), Ket(0.27), Baldi(0.16).
38	Decanoic acid ethyl ester	22.262	5	Fuss(1.44), Baladi(1.3), Alfons(1.26), Succari(0.600), Naomee(0.46).
39	Hexadecanoic acid ethyl ester	36.065	5	Langary(1.07), Naomee(0.99), Alfons(0.700), Misk(0.25), Tomy(0.19).
40	Tetradecanoic acid ethyl ester	31.899	4	Naomee(4.61), Alfons(1.83), Langary(0.68),

DOI: 10.9790/3008-10217782

www.iosrjournals.org

# GC-MS Study on the Aroma of Thirteen Egyptian Mango Cultivars

				Baladi(0.45).
41	Delta-cadinene	25.706	4	Baladi(0.57), Misk(0.55), Langary(0.54), Ket(0.17).
42	Gamma-terpinene	12.229	4	Tomy(0.33), Fagr(0.26), Ket(0.24), Misk(0.15).
43	Hexanoic acid ethyl ester	10.396	4	Zebda(1.18), Alfons(0.85)Baladi(0.34), Langary(0.34)
44	Ethandionediphenyl	32.50	4	Alfons(0.72), Ket(0.60), Misk(0.21), Baladi(0.20).
45	3-nonen-2-one	14.828	3	Misk(1.09), Zebda(1.00), Baladi(0.51),
46	3(2H)-furanone, dihydro-2- methyl	4.784	3	Succari1.06), Fagr(0.46), Misk(0.15).
47	Dodecanoic acid ethyl ester	27.327	3	Naomee(1.05), Baladi(0.93), Alfons(0.69).
48	Benzene , methyl(1- methylethyl)	11.122	3	Tomy(0.47), Ket(0.28), Langary(0.27).
49	2,3-dihydro-3,5-dihydroxy-6- dimethyl -4H-pyran-4-one	14.941	3	Fuss(1.93), Langary(0.31), Ket(0.16).
50	Ethyl-2-octaneoate	18.027	3	Fuss(1.97), Baladi(0.27), Langary(0.27).
51	Alpha-seliene	25.021	3	Misk(0.64), langary(0.33)), Alfons(0.16).
52	(+)-2-carene	10.892	2	Zebda(2.23), Ewaise(1.42)
53	Santene(Norbormene)	15.663	2	Fuss(1.92), Tomy(0.12).
54	Mesifurane	12.351	2	Misk(1.25), Zebda(0.81).
55	5,7-octadien-2-ol,2,6-dimethyl	15.224	2	Fagr(0.56), Soba el-set(0.19).
56	1,8-menthadien-4-ol	15.975	2	Fuss(2.85), Baladi(0.09).
57	2,4-nonadiene	18.348	2	Alfons(0.98), Succari(0.28).
58	Azulene	25.111	2	Misk(0.75), Baladi(0.12).
59	Tridecanal	25.246	2	Ket(0.74), Baladi(0.38).
60	Delta-guaiene	25.272	2	Misk(2.25), Langary(0.28).
61	Dodecanoic acid	26.557	2	Fagr(1.13), Alfons(0.11).
62	1-decanol	16.947	1	Alfons(5.37).
63	1-dodecanol	24.024	1	Alfons(1.61).
64	Ethyl-4-deceneoate	21.124	1	Alfons(1.69).
65	Formic acid isobornyl ester	20.786	1	Alfons(5.98).
66	Methyl decadienoate	21.195	1	Alfons(1.18).
67	Nonanoic acid ethyl ester	12.447	1	Alfons(31.29).
68	Undecanoic acid ethyl ester	27.221	1	Alfons(2.03).
69	Ethyl tiglate	8.489	1	Baladi(2.34).
70	Octane	4.488	1	Baladi(0.74).
71	7-ethylidene-6b,7,8,8a- tetrahydrocylobut acenaphythalene.	5.097	1	Ewaise(1.65).
72	(-)-isosativene	23.057	1	Ewaise(1.96).
73	Sylvestrene	11.286	1	Ewaise(1.34).
74	2H-pyran,2-ethenyl tetrahydro-2,6,6-trimethyl	9.443	1	Fagr(2.63).
75	Ocimenyl acetate	15.558	1	Fagr(0.68).
76	Benzene methanol,4-(1- methylethyl)	16.23	1	Fuss(2.60).
77	P-menth-1,5,8-triene	13.912	1	Fuss(2.34).
78	2-cyclopentene,1,4-dione	6.856	1	Succari(0.91).
79	2-furanmethanol	6.073	1	Succari(3.10).
80	Henicosan,11-(1-ethylpopyl)	40.695	1	Succari(2.38).
81	Hexatriacontane	39.515	1	Succari(1.06).
82	1-octadecene	40.331	1	Succari(4.69).
83	2-Beta pinene	9.577	1	Tomy(1.69).
84	(+)-mentha-1,8-diene	13.11	1	Tomy(0.33).

### IV. Conclusion

### From the above table, one can conclude that:

Beta-Myrcene is common in all cultivars except Ket, while 2-Furan-carboxaldehyde and Ethanone, 2,2dimethoxy-1,2 diphenyl, are common in all cultivars except Fuss. So, it can be concluded that these three components are responsible for the general characteristic fragrance of Mango fruits.

On the other hand, each cultivar has its own unique and characteristic fragrance due to components present solely in its essential oil; together with the presence or absence of other components in variable percentages.

From the above mentioned statements, one can identify the mango fruit in general and its cultivar in particular. Also, this data will help to prepare artificial Mango flavors which are needed in pharmaceutical, food and cosmetics industries. This is shown as follow:

Alfons cultivar has the highest percentage of the common components: Butanoic acid ethyl ester, 1-heptadecanol, Ethandionediphenyl and 2,4-nonadiene. And has variable percentage of other common components. But it has its unique components, namely: 1-decanol(5.37%), 1-dodecanol(1.61%), ethyl-4-

deceneoate(1.69%), formic acid isobornyl ester(5.98%), methyldecadienoate(1.18%), nonanoic acid ethyl ester(31.29%), and undecanoic acid ethyl ester(2.03%).

**Baladi cultivar** has the highest percentage of the common components: Beta-myrcene, nonanal, and alphacadinene. And has variable percentage of other common components. But it has its unique components: ethyl tiglate(2.34%)and octane(0.74%).

**Ewaise cultivar** has the highest percentage of the common components: Linalool oxide, germacrene-D, and benzene acetaldehyde. And has variable percentage of other common components. But it has its unique components: 7-ethylidene-6b,7,8,8a-tetrahydrocyclobut-acenaphytalene(1.65%), (-)-isosativene(1.96%), and sylvestrene(1.34%).

**Fagr cultivar** has the highest percentage of the common components: Alpha-terpineol, Linalool, 5,7-octadien-2-ol,2,6-dimethyl, and dodecanoic acid. And has variable percentage of other common components. But it has its unique components: 2H-pyran,2-ethenyl tetrhydro-2,6,6-trimethyl(2.63%), and ocimenyl acetate(0.68%).

**Fuss cultivar** has the highest percentage of the common components: Beta-citronellol, geraneol, Alphaterpinene, 2-butenoic acid ethyl ester, borneol, decanoic acid ethyl ester, 2,3-dihydro-3,5-dihydroxy-6-dimethyl-4H-pyran-4-one, ethyl-2-octaneoate, Santene, and 1,8-menthadien-4-ol. And has variable percentage of other common components. But it has its unique components: Benzene methanol,4-(1-methylethyl)(2.60%),and Pmenth-1,5,8-triene(2.34%).

**Ket cultivar** has the highest percentage of the common components: Ethanone,2,2-dimethoxy-1,2-diphenyl, hexadecanal, and Camphene. And has variable percentage of other common components. But it has no unique components.

**Langary cultivar** has the highest percentage of the common components: 2-heptadecanone, 2-propenoic acid, 2-methylethyl ester, 2-propanone-1-hydroxy, and hexadecanoicacid ethyl ester. And has variable percentage of other common components. But it has no unique components.

**Misk cultivar** has the highest percentage of the common components: trans-caryophyllene, Alpha-humulene, 3nonen-2-one, Alpha-seliene, Mesifurane, Azulene, and Delta-guaiene. And has variable percentage of other common components. But it has no unique components.

**Naomee cultivar** has the highest percentage of the common components: Alpha-copaene, tetradecanoic acid ethyl ester, and Dodecanoic acid ethyl ester. And has variable percentage of other common components. But it has no unique components.

**Sobaa El-set cultivar** has the highest percentage of the common components: 1,3,6-octatriene,3,7-dimethyl(E) and cis-ocimene. And has variable percentage of other common components. But it has no unique components.

**Succari cultivar** has the highest percentage of the common components: 2-furancarboxaldehyde, 2-furancarboxaldehyde,5-methyl, Ethanone,1-(2-furanyl), and Alpha-cedrol, 3(2H)-furanone, dihydro-2-methyl . And has variable percentage of other common components. But it has its unique components: 2-cyclopentene,1,4-dione(0.91%), 2-furanmethanol(3.10%), Henicosan,11-(1-ethylpropyl)(2.38%), Hexatriacontane(1.06%), and 1-octadecene(4.69%).

**Tomy cultivar** has the highest percentage of the common components: Delta-3-carene, Alpha-pinene, dl-limonene, Gamma-terpinene, and Benzene, methyl (1-methylethyl). And has variable percentage of other common components. But it has its unique components: Beta-pinene(1.69%) and (+)-mentha-1,8-diene.

**Zebda cultivar** has the highest percentage of the common components: Alpha-terpinolene, cis-methyl propenylketon, Octanoic acid ethyl ester, l-phellandrene, N-heptanal, Hexanoic acid ethyl ester, and (+)-2-carene. And has variable percentage of other common components. But it has no unique components.

### Acknowledgement

The authors are grateful to Arab Company for Pharmaceutical and Medicinal Plants "Mepaco-Medifood" for her ultimate support for research and development team.

### References

- H.Edurado, S. Ramos, et al. Chemical composition, Leishmanicidal and Cytotoxic Activities of the Essential oils from [1]. MangiferaindicaLvar. Rosa and Espada, J.BioMed Research International, Vol.2014, 2014.
- [2]. U.P. De Albuquerque, et al. Medicinal Plants of the caatinga(semi-arid) Brazil: A Quantitative Approach, J. of Ethnopharmacology, vol.114, No.3, pp.325-354, 2007.
- [3]. Seham S. El-Hawary and Mohamed A. Rabeh, Mangiferaindica peels: A common waste product with impressive immunostimulant, anticancer and antimicrobial potency, J. of Natural Sciences Research, Vol. 4, No. 4, 2014.
- [4]. Robert J. Knight,"Report on the Egyptian Mango Industry", Florida Agricultural Experiment Station, J. series No. N-01179, 2014.
- A.Eloisa Helena and Andrade etal. Aroma Volatile Constituents of Brazilian Varieties of Mango Fruit, J. of Food Composition and [5]. Analysis, vol. 13, pp.27-33, 2000.
- [6]. J.C. Medina. FrutasTropicais-Manga, ITAL, Sao Paulo, 1981.
- [7]. L. El-Tomi. Subtropical fruit industry in Egypt, Proc. Fla. State Hort. Soc. 66: 195-198, 1953.
- [8]. R.J. Knight.Evaluating important fruit characters in Mango germplasm, Fruit var. J. 47(1): 25-30, 1993.
- L. B. Singh. The Mango, Interscience Publishers, New York, 438 illus. Map. 1960. S. M. Wheatly. "The Mangoes of the Kenya Coast E. Afr. Agric., J. 22: 46-54, 1956. [9].
- [10].